

# THERMAL BIOLOGY OF A POPULATION OF *XENOSAURUS NEWMANORUM* (SQUAMATA: XENOSAURIDAE) FROM XILITLA, SAN LUIS POTOSÍ, MEXICO: DO THEY ACTIVELY THERMOREGULATE?

## BIOLOGÍA TÉRMICA DE UNA POBLACIÓN DE *XENOSAURUS NEWMANORUM* (SQUAMATA: XENOSAURIDAE) DE XILITLA, SAN LUIS POTOSÍ, MÉXICO: ¿TERMORREGULAN ACTIVAMENTE?

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**Resumen.**— Las lagartijas de escamas granulares (familia Xenosauridae) comprenden un clado de escamados norteamericanos especializados a la vida en grietas, con una tendencia hacia temperaturas corporales bajas y altas tasas de pérdida de agua, y son a menudo consideradas termoconformistas. No obstante, existen pocos estudios sobre la ecología térmica de estas lagartijas, la mayoría de estos limitados a documentar las temperaturas corporales y su relación con las temperaturas ambientales. En este trabajo, documentamos las temperaturas corporales en campo ( $T_b$ ), las temperaturas seleccionadas en condiciones de laboratorio ( $T_{set}$ ), las temperaturas operativas ( $T_o$ ), la eficiencia en la termorregulación y las tolerancias térmicas de 10 individuos de Lagartijas de Escamas Granulares de Newman, *Xenosaurus newmanorum*, recolectados en otoño de 2017 en la región de La Huasteca Potosina, México. El promedio de  $T_b$  fue de 22.75 °C y el de  $T_{set}$  de 25.08 °C (1.64 °C y 3.97 °C por encima del promedio de  $T_o$ , respectivamente). La especie en cuestión tuvo un intervalo de tolerancia térmica relativamente estrecho (25.89 °C). Además, con base en dos índices de termorregulación, encontramos evidencia directa de termorregulación activa en *X. newmanorum*. En este estudio documentamos el primer registro de termorregulación activa en un xenosáurido de un entorno de baja elevación y el segundo registro de tolerancias térmicas. Aunque se requiere de trabajo de campo y laboratorio más exhaustivos a fin de verificar este patrón, nuestros resultados son consistentes con estudios previos y sugieren que la termorregulación activa podría estar más extendida en el género de lo que previamente se suponía.

**Palabras clave.**— temperatura corporal seleccionada, temperatura máxima voluntaria, termorregulación activa, tolerancia térmica crítica, Xenosauridae.

**Abstract.**— The knob-scaled lizards (family Xenosauridae) comprise a clade of crevice-dwelling North American lizards with a trend towards low body temperature and high rates of water loss, and are often regarded as thermoconformers. However, there are few studies on their thermal ecology, most of them limited to documenting the body temperatures and their relation with environmental temperatures. Here, we documented the field body temperatures ( $T_b$ ), selected temperatures ( $T_{set}$ ), operative temperatures ( $T_e$ ), thermoregulatory effectiveness, and thermal tolerances of 10 individuals of Newman's Knob-scaled Lizards, *Xenosaurus newmanorum*, collected in the fall of 2017 in La Huasteca region of Mexico. Mean  $T_b$  was 22.75 °C and mean  $T_{set}$  25.08 °C (1.64 °C and 3.97 °C above mean  $T_e$ , respectively). The species had a relatively narrow thermal tolerance range (25.89 °C). Furthermore, based on two different thermoregulatory indexes, we found direct evidence of active thermoregulation in this sample of *X. newmanorum*. This constitutes the first record of active thermoregulation in a xenosaurid from a low-elevation environment and the second record of thermal tolerances. Although more extensive field and laboratory work is needed to verify this pattern, our results are consistent with previous studies suggesting that active thermoregulation might be more widespread in the genus than previously thought.

**Keywords.**— active thermoregulation, critical thermal tolerances, selected body temperature, voluntary thermal maximum, Xenosauridae.

## INTRODUCTION

Lizards of the genus *Xenosaurus* are characterized by their flattened morphology, crevice-dwelling habits, reduced distributional ranges, and low vagility (Zamora-Abrego & Ortega-León, 2016). Most of the 18 currently recognized species (12 described + six undescribed taxa; Nieto-Montes de Oca et al., 2017) are found in densely covered habitats, such as cloud forests and tropical rainforests, where canopy tend to limit the incidence of solar radiation and thus the opportunities for thermal regulation (Berriozabal-Islas et al., 2018). The genus *Xenosaurus* is considered a practically tigmotherm and thermoconformer clade with a trend towards low thermal body temperatures (Lemos-Espinal et al., 1998; Vitt & Caldwell, 2014; García-Rico et al., 2015). However, some species in the genus (e.g. *X. phalaroanthereon*, *X. mendozai*, and *X. rectocollaris*) are known to present differences between body and environmental temperatures (Lemos-Espinal et al., 1996; Lemos-Espinal et al., 2004; Lemos-Espinal & Smith, 2005; Woolrich-Piña et al., 2012), which may reflect the thermal pressures exerted by the environment, as these lizards occur in high elevation habitats with low canopy coverage (Lemos-Espinal & Smith, 2005; Woolrich-Piña et al., 2012). These thermally heterogeneous habitats may provide greater thermoregulation opportunities because they offer a wide range of temperatures to choose at microhabitat level (Hertz et al., 1993).

At this time, most studies on the thermoregulation of members of the genus *Xenosaurus* have limited to characterize their body temperatures and its relation with environmental temperatures, while the thermal tolerances of these lizards remain largely unstudied (Clusella-Trullas & Chown, 2013; Berriozabal-Islas, 2018). Likewise, effectiveness of thermoregulation has been

properly evaluated only for two species of knob-scaled lizards (Woolrich-Piña et al., 2012; Berriozabal-Islas, 2018). Given that thermal traits such as selected temperatures, thermal tolerances and the degree of thermoconformity or thermoregulation have direct implications on the vulnerability of ectotherms to habitat modification and climate change (Nowakowski et al., 2018; Arenas-Moreno et al., 2021), their quantification might provide valuable clues to the assessment of extinction risk, particularly for threatened and range-restricted species, as is the case of most species of *Xenosaurus* (Wilson et al., 2013; García-Padilla et al., 2020).

Newman's Knob-scaled Lizards (*Xenosaurus newmanorum*) are crevice specialist lizards whose distribution is restricted to the areas with subtropical and secondary vegetation (coffee and citrus plantations) of Xilitla, San Luis Potosí, and Pisaflores, Hidalgo, at elevations between 390 and 880 m a.s.l. (Lemos-Espinal et al., 1998; Lemos-Espinal et al., 2012; Lara-Tufiño et al., 2013). As well as most of its congeners, this species inhabits very shaded environments, where achievement of high temperatures might be difficult (Lemos-Espinal et al., 1998). Also, it has been shown to present relatively high rates of evaporative water loss, compared to other mesic environment lizards (Muñoz-Nolasco et al., 2019). This species is listed as endangered according to the International Union for Conservation of Nature (IUCN; Lavin & Mendoza, 2007), under special protection in the Mexican Endangered Species Act (NOM-059-SEMARNAT-2010 and its 2019 amendment to the Normative Appendix III; DOF, 2010, 2019), and is placed in the high vulnerability category according to Wilson et al. (2013), with an Environmental Vulnerability Score of 15. Lemos-Espinal et al. (1998) already provided data on the thermal ecology of this species (body and microhabitat temperatures) from the area of Xilitla, San Luis

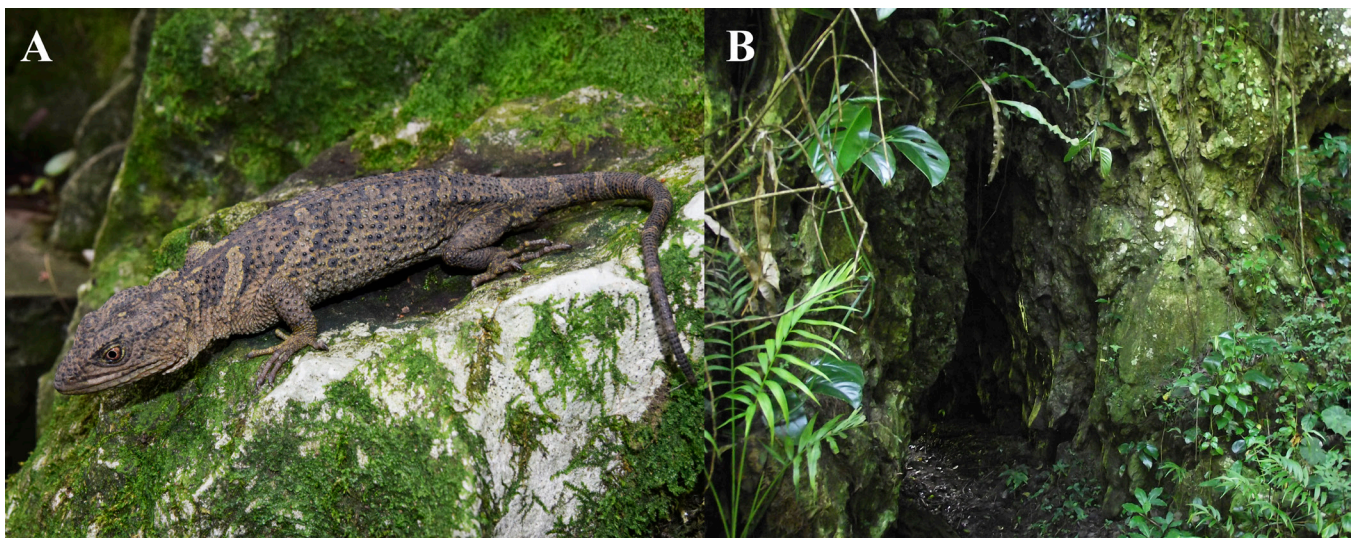
Potosí. More recently, Berriozabal-Islas (2018) calculated some thermoregulatory indexes for another population at the locality of Pisaflores, Hidalgo, finding a thermoconformer pattern. Here, we studied a small sample of *X. newmanorum* from Xilitla, San Luis Potosí, to test whether the species might be able to actively thermoregulate and to determine its thermal tolerances (critical thermal minimum and maximum; voluntary thermal maximum). Given the dense canopy cover of its habitat that limits solar radiation and hence produces relatively low and stable temperatures (Lemos-Espinal et al., 1998), we expected *X. newmanorum* to show some degree of active thermoregulation and to be stenothermal (i.e. have a narrow tolerance range).

## MATERIALS AND METHODS

From 8 to 13 October 2017, we collected 10 individuals (four males and six females) of *X. newmanorum* (mean snout-vent length = 100.27 mm, SD = 13.59 mm, 62.9 – 110.6 mm) in an area of secondary tropical evergreen forest and coffee plantations in the municipality of Xilitla, San Luis Potosí, Mexico (21.380° N, 98.987° W, 687 m elev.; Fig. 1). Mean annual temperature at the study site is 22.5 °C, with a maximum temperature of the warmest month of 33.3 °C, a minimum temperature of the coldest month of 10.4 °C, and a mean annual precipitation of 2260 mm (data obtained through WorldClim version 2 at 2.5' resolution; Fick & Hijmans, 2017). We caught only active individuals (as determined by exposure of their heads outside the crevices) aided by a malleable metal wire with which we gently removed lizards from their crevices. As we did not take more than 60 seconds in removing

each lizard, and given that we did not directly handle them when inside the crevice, it is unlikely that capture has influenced the body temperature of the organisms. Immediately after capturing the lizards, we recorded their body (cloacal) temperature ( $T_b$ ) with a digital readout thermometer Fluke® 51-II with a type T thermocouple ( $\pm 0.1$  °C), air temperature ( $T_a$ ) at 3 cm above substrate, and substrate temperature ( $T_s$ ) inside the crevice at the depth at which we first spotted the organism (Woolrich-Piña et al., 2006). We determined the relationship between  $T_b$  and  $T_a$  and  $T_s$  through simple linear regressions (Lemos-Espinal et al., 1998; García-Rico et al., 2015; Cardona-Botero et al., 2019).

We also recorded environmental operative temperatures ( $T_e$ ), which are the temperatures that organisms would experience in the field in the absence of thermoregulatory behavior (Bakken, 1992; Dzialowski, 2005). In order to do that, we first performed a calibration of models intended to emulate the rates of heat gain and loss. For this, we subjected an adult individual (immobilized with medical tape) along with a set of four candidate models made of polyvinyl chloride (PVC) pipes varying in shape, size, and color to alternating 10-minutes periods of artificial incandescent light and shade for an hour in a lab setting (Arenas-Moreno et al., 2021). We recorded temperatures every two seconds using an Omega TC-8 data logger with T-type ultrathin thermocouples, one affixed with medical tape to the belly of the lizard and other four connected to the inside of each of the candidate models. Then, we performed a simple linear regression using the temperature recordings of each candidate model against the temperature of the lizard and chose the model with the highest



**Figura 1.** Individuo de *Xenosaurus newmanorum* (A) y su hábitat en Xilitla, San Luis Potosí, México (B). Fotos: Adán Bautista-del Moral.

**Figure 1.** Individual of *Xenosaurus newmanorum* (A) and its habitat in Xilitla, San Luis Potosí, Mexico (B). Photos: Adán Bautista-del Moral.



significant determination coefficient (Bakken, 1992; Dzialowski, 2005). The best model ( $r^2 = 0.87$ ,  $F_1$ ,  $1390 = 9652.8$ ,  $P < 0.001$ ) consisted of a heat-flattened, gray-painted pipe 7 cm length, 2.7 cm width, and 1.5 cm height. We coupled two calibrated models to each of three data loggers HOBO® U12-006 and placed them at different levels of exposure in crevices where we previously found individuals of *X. newmanorum*. We set data loggers to record  $T_c$  every 10 minutes along the observed activity period of the species (09:00 to 17:00 h) during the five days of fieldwork (~40 h).

We transported the collected lizards to proper facilities near the study site to continue with the experiments and placed them in individual plastic terraria (16 × 16 × 7 cm) with peat moss and rocks for two days prior to the onset of the experimental procedure, with no food but access to water ad libitum (Lara-Resendiz et al., 2013). To measure the selected temperatures ( $T_{set}$ ), we build a thermal gradient made of panels of cellular polycarbonate (130 × 100 × 50 cm), at the bottom of which we scattered peat moss and stacked natural flagstones from the study site, in order to provide shelter along the gradient and emulate habitat conditions. At one extreme of the enclosure, we suspended four 100 W incandescent lightbulbs 30 cm above it, and at the other extreme we placed refrigerating gel bags, providing a gradient of temperatures ranging from ~10 to 38 °C (Arenas-Moreno et al., 2018, 2020). Although knob-scaled lizards are primarily tigmothermal and hence exposure to direct light might affect the selection of temperatures inside the gradient, the arrangement of the flagstones limited the incidence of artificial light while allowing their heating at that extreme. We carried out a gradient experiment the day following the capture of the individuals, registering  $T_{set}$  at one-hour intervals from 09:00 to 17:00 h (the observed daily activity period of the species; pers. obs.) with one hour of previous habituation (Lara-Resendiz et al., 2013; Arenas-Moreno et al., 2018; Caetano et al., 2020).

From the data of  $T_b$ ,  $T_c$ , and the first and third quartiles of the  $T_{set}$  data ( $T_{set}$  range), we calculated the accuracy of thermoregulation ( $db$ ), the thermal quality of habitat ( $de$ ), and the effectiveness of thermoregulation indexes proposed by Hertz et al. (1993) [ $E = 1 - (db/de)$ ] and Blouin-Demers & Weatherhead (2001) ( $de - db$ ). Low values of  $db$  and  $de$  indicate high accuracy of thermoregulation and high thermal quality of the habitat, respectively, while high values indicate the opposite. In the case of the thermoregulatory effectiveness indexes ( $E$  and  $de - db$ ), positive values represent active thermoregulation, zero represents perfect thermoconformity, and negative values, a suboptimal exploitation of the thermal offer. However, whereas the Hertz et al. (1993) index is relative and goes from

0 to 1, the Blouin-Demers & Weatherhead (2001) index is an open ended and absolute measure (in °C) of the extent of deviation from thermoconformity. Although the Blouin-Demers & Weatherhead (2001) has been regarded as a more reliable measure of thermoregulatory efficiency, we also provide the Hertz et al. (1993) index due to its widespread use (Taylor et al., 2020). To assess the thermal tolerance of the species, we recorded the critical thermal minimum and maximum ( $CT_{min}$  and  $CT_{max}$ , respectively), which refer to the lowest and highest temperatures at which organisms lose their locomotor capabilities (loss of righting response within 10 s after placing them on their dorsum; Kaufmann & Bennett, 1989), as well as the voluntary thermal maximum ( $VT_{max}$ ), which, as its name indicates, is defined as the highest temperature at which they avoid further heating (Carothers et al., 1997; Camacho et al., 2019). Unfortunately, we were not able to measure voluntary thermal minimum due to the lack of proper equipment. For the estimation of  $CT_{min}$ , we placed the lizards in a plastic container and this, in turn, in an ice-filled box, to gradually reduce its body temperature.

For the estimation of  $VT_{max}$ , we placed each individual ( $T_b$  ~24 °C) in a transparent, uncovered plastic container for ~3 min until it remained calm. Then, we turned on a 150 W light bulb suspended 30 cm above the container and watched carefully the lizard at a 2 m distance until it started trying to evade the heat source and escaping from the container, moment at which we recorded its  $T_b$  as  $VT_{max}$ . Immediately after this, we returned the individual to the same container to keep gradually heating it until reaching its  $CT_{max}$ , based on the same criterion of loss of righting response (Arenas-Moreno et al., 2021). Organisms were promptly and gradually brought closer to their  $T_{set}$  after reaching their critical thermal points, so that no permanent negative effect was inflicted to them. We carried out the thermal tolerance experiments on the day following the gradient experiment and similarly performed the cold tolerance ( $CT_{min}$ ) and heat tolerance ( $VT_{max}$  and  $CT_{max}$ ) experiments ~24 h apart, with half the sample being subjected first to the cold tolerance and the other half to the heat tolerance experiments. After finishing the experimental procedure, we verified all the organisms have recovered from the tests and then released them back to their respective crevices in the field.

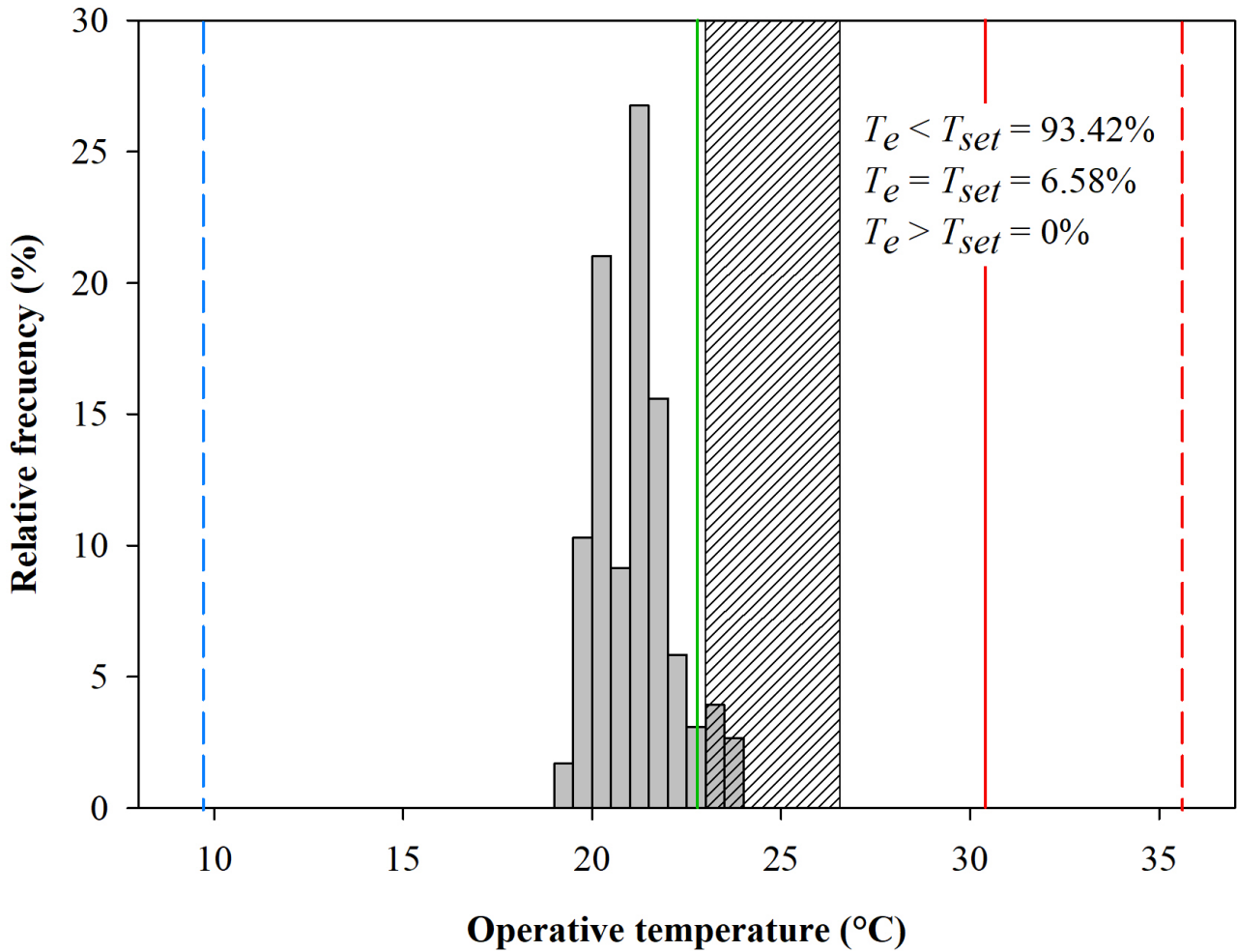
## RESULTS

The thermal traits of the sample of *X. newmanorum* here documented are summarized in Table 1, along with traits of other members of the genus documented in the literature. Throughout the text, values are presented as mean ± standard error (minimum value – maximum value). For the organisms

**Tabla 1.** Temperaturas corporales en campo ( $T_b$ ), temperaturas seleccionadas (promedio de  $T_{set}$  y  $T_{set}$  25-75%) y eficiencia en la termorregulación ( $E$ ; sensu Hertz et al., 1993, y  $de - db$ , sensu Blouin-Demers & Weatherhead, 2001) de las especies de *Xenosaurus* documentadas en la literatura. Se muestran también el tipo de vegetación (Veg.) y la altitud (Elev.) en cada localidad. La información de *X. newmanorum* aquí documentada se indica en negritas. ND = sin datos. Tipo de vegetación: AP = plantaciones agrícolas; CF: bosque mesófilo; OF = bosque de encino; PF = bosque de pino; POF = bosque de pino-encino; TDF = bosque tropical caducifolio; TEF = bosque tropical perennifolio; XS = matorral xerófilo. Los datos de *X. mendozai* de Lemos-Espinal (2004) fueron referidos previamente como *X. platyceps* y luego reasignados de acuerdo con Nieto-Montes de Oca et al. (2013). Los valores de temperatura se presentan como promedio  $\pm$  EE. El signo de interrogación (?) representa información no especificada.

**Table 2.** Field body temperatures ( $T_b$ ), selected body temperatures (mean  $T_{set}$  and  $T_{set}$  25-75%) and effectiveness of thermoregulation ( $E$ , sensu Hertz et al., 1993, and  $de - db$ , sensu Blouin-Demers & Weatherhead, 2001) of the species of *Xenosaurus* documented in the literature. Vegetation type (Veg.) and elevation (Elev.) at each locality is also shown. Data of *X. newmanorum* herein documented is indicated in bolds. ND = no data. Vegetation type: AP = agricultural plantations; CF: cloud forest; OF = oak forest; PF = pine forest; POF = pine-oak forest; TDF = tropical dry forest; TEF = tropical evergreen forest; XS = xerophilous scrub. Data of *X. mendozai* from Lemos-Espinal et al. (2004) was formerly referred as *X. platyceps* and then reassigned according to Nieto-Montes de Oca et al. (2013). Temperature values are presented as mean  $\pm$  SE. Question mark (?) represents non-specified data.

Species	$T_b$ (°C)	$T_{set}$ (°C)	$T_{set}$ 25-75% (°C)	Veg.	Elev. (m)	$E$	$de - db$ (°C)	References
<i>X. agrenon</i>	25.6 $\pm$ 0.3	ND	ND	TDF/OF/ PF/ POF	1000 - 1470	ND	ND	Lemos-Espinal et al. (2003)
<i>X. fractus</i>	19.12 $\pm$ 0.53	ND	ND	POF	1900 - 2170	ND	ND	Cardona-Botero et al. (2019)
<i>X. fractus</i>	19.67 $\pm$ 0.39	21.69	ND	CF/POF	1700	ND	ND	Woolrich-Piña et al. (2020)
<i>X. grandis</i>	21.4 $\pm$ 0.19	ND	ND	CF/AP	1050 - 1150	ND	ND	Cardona-Botero et al. (2019)
<i>X. mendozai</i>	20.6 $\pm$ 0.4	ND	ND	OF	1180	ND	ND	Lemos-Espinal et al. (2004)
<i>X. mendozai</i>	23.39	22.18	?	?	?	0	-0.72	Berriozabal-Islas (2018)
<i>X. newmanorum</i>	22.9 $\pm$ 0.2	ND	ND	TEF/AP	ND	ND	ND	Lemos-Espinal et al. (1998)
<i>X. newmanorum</i>	21.6 $\pm$ 0.3	ND	ND	TEF	389	ND	ND	Lara-Tufiño et al. (2013)
<i>X. newmanorum</i>	19.3 $\pm$ 0.9	ND	ND	AP/TEF	825	ND	ND	Lara-Tufiño et al. (2013)
<i>X. newmanorum</i>	24.8 $\pm$ 0.3	ND	ND	AP/TEF	880	ND	ND	Lara-Tufiño et al. (2013)
<i>X. newmanorum</i>	23	24.65	?	TEF	?	0.4	ND	Berriozabal-Islas (2018)
<i>X. newmanorum</i>	22.75 $\pm$ 0.43	25.08 $\pm$ 0.290	23 - 26.6	TEF/AP	687	0.68	1.3	This study
<i>X. phalaroanthereon</i>	20.3 $\pm$ 0.4	ND	ND	OF	2185	ND	ND	Lemos-Espinal & Smith (2005)
<i>X. rectocollaris</i>	23.2 $\pm$ 0.3	30.4	26 - 33.4	XS	2100 - 2400	0.52 (0.42 - 0.62)	ND	Woolrich-Piña et al. (2012)
<i>X. tzacualtipantecus</i>	18.09 $\pm$ 0.24	ND	ND	TEF	1900	ND	ND	García-Rico et al. (2015)
<i>X. tzacualtipantecus</i>	18.26	24.06	?	?	?	0.4	2.83	Berriozabal-Islas (2018)



**Figura 2.** Distribución de las temperaturas operativas ( $T_e$ ) de *Xenosaurus newmanorum* en su hábitat en Xilitla, San Luis Potosí. El área sombreada representa el intervalo de temperaturas seleccionadas ( $T_{set}$  25-75%); la línea verde, el promedio de temperatura corporal en campo ( $T_b$ ); la línea azul discontinua, la temperatura crítica mínima ( $CT_{min}$ ); la línea roja continua, la temperatura máxima voluntaria ( $VT_{max}$ ), y la línea roja discontinua, la temperatura crítica máxima ( $CT_{max}$ ). Se muestra también el porcentaje de temperaturas operativas por debajo, dentro y por encima de  $T_{set}$  25-75%.

**Figure 2.** Distribution of operative temperatures ( $T_e$ ) of *Xenosaurus newmanorum* in its habitat at Xilitla, San Luis Potosí. Shaded area represents the range of selected temperatures ( $T_{set}$  25-75%); green line, the mean field body temperature ( $T_b$ ); blue dashed line, the critical thermal minimum ( $CT_{min}$ ); solid red line, the voluntary thermal maximum ( $VT_{max}$ ), and red dashed line, the critical thermal maximum ( $CT_{max}$ ). It is also showed the percentage of  $T_e$  below, within, and above  $T_{set}$  25-75%.

of the population studied,  $T_b$  was lower than  $T_{set}$  and showed no significant relationship with  $T_a$  ( $F_{1,7} = 3.457, r^2 = 0.331, P = 0.105$ ) nor with  $T_s$  ( $F_{1,7} = 5.375, r^2 = 0.434, P = 0.054$ ). Most of the  $T_e$  recorded (93.42%) were below the  $T_{set}$  range, while only 6.58% of  $T_e$  matched the  $T_{set}$  range (Fig. 2). Consistent with this, *X. newmanorum* showed a relatively high accuracy of thermoregulation ( $db = 0.61$  °C) and a moderately low thermal quality of the habitat ( $de = 1.91$  °C), which together indicated active thermoregulation according to both thermoregulatory effectiveness indexes used ( $E = 0.68$ ;  $de - db = 1.3$  °C). The thermal tolerance range ( $CT_{max} - CT_{min}$ ) of the species was 25.89 °C, with a  $CT_{min}$  of  $9.78 \pm 0.45$  °C (7.2 – 11.4

°C), a  $VT_{max}$  of  $30.39 \pm 0.41$  °C (28.7 – 31.9 °C), and a  $CT_{max}$  of  $35.67 \pm 0.18$  °C (34.9 – 36.8 °C).

## DISCUSSION

*Xenosaurus newmanorum* exhibited a  $T_b$  similar to that of other species of *Xenosaurus* from tropical habitats (Table 1). However, conversely to what Lemos-Espinal et al. (1998) found,  $T_b$  was not significantly related to  $T_a$  nor  $T_s$ . Although this may be due to the small sample size of our study, it also may reflect active thermoregulation, but, as discussed below, cannot be regarded

per se as a conclusive proof of this pattern. Also, its  $T_b$  and  $T_{set}$  are consistent with previous data for this species (Lemos-Espinal et al., 1998; Berriozabal-Islas, 2018). Among species of the genus, *X. fractus* and *X. rectocollaris* have the lowest and highest  $T_{set}$ , respectively (Woolrich-Piña et al., 2012; Woolrich-Piña et al., 2020), with *X. newmanorum* being halfway on this parameter.

Based on the scarce information available on the thermal ecology of knob-scaled lizards, it seems that most *Xenosaurus* species live in environments that offer temperatures below their  $T_{set}$  range (Woolrich-Piña et al., 2012; Berriozabal-Islas, 2018). In other crevice-dwelling lizards, such as some members of the genera *Lepidophyma* and *Xantusia*, a low thermal quality of the habitat has been linked to a shift towards active thermoregulation (Arenas-Moreno et al., 2018, 2021). Therefore, our results suggesting active thermoregulation in *X. newmanorum* should not be unexpected but just a pattern consistent with the cost-benefit model of thermoregulation (Huey & Slatkin, 1976).

Previous studies claiming to find support for a thermoconformer pattern in *Xenosaurus* have relied on the relationship between field body and microhabitat temperatures (e. g. Lemos-Espinal et al., 1998; García-Rico et al., 2015; Cardona-Botero et al., 2019). However, this approach has been proven not to be a proper measure of where on the thermoconformer – thermoregulating continuum an ectotherm sits, as other biophysical factors, such as solar radiation, might influence body temperature, and as the relationship between body and microhabitat temperatures is non-linear (Dreisig, 1984; Hertz et al., 1993). Nonetheless, comparisons between body and microhabitat temperatures, or even just body temperatures, have provided indirect indications for active thermoregulation in at least two species: *X. phalaroanthereon*, which is known to keep a  $T_b \approx 5^\circ\text{C}$  above  $T_a$  and  $T_s$  (Lemos-Espinal & Smith, 2005), and *X. rectocollaris* in which pregnant females have higher body temperatures than non-pregnant females (Woolrich-Piña et al., 2012). These two species inhabit high elevation (>2000 m) thermally heterogeneous habitats, where low temperatures make thermoregulation an advantageous strategy. *Xenosaurus newmanorum*, on the other hand, is a lowland species, and hence it would be expected to be exposed to more favorable temperatures and to follow a thermoconformer pattern. But as our data shows, this species is mostly subjected to environmental temperatures below its  $T_{set}$  range, probably due to the dense canopy cover of its habitat, making active thermoregulation a more convenient strategy.

Even the use of just one thermoregulatory index might yield a partial perspective of the thermal ecology of a given species,

and could also lead to overlook proofs of active thermoregulation in seemingly non-thermoregulating ectotherms. For instance, Berriozabal-Islas (2018) calculated the Hertz et al. (1993; *E*) index for three species of *Xenosaurus*, finding a thermoconformer strategy. However, based on the *db* and *de* values provided by the author, and according to the Blouin-Demers and Weatherhead (2001; *de - db*) index, *X. newmanorum* (from Pisaflores) and *X. tzacuaultipantecus* are both active and effective thermoregulators (Table 1). Given that the *E* index has been criticized for its mathematical assumptions and interpretability (Blouin-Demers & Weatherhead, 2001), the *de - db* index should be taken as a more conclusive measure of thermoregulation. Because of the afore mentioned, it seems plausible that active thermoregulation is more widespread than previously thought in *Xenosaurus* lizards.

Our study is the second one to document thermal tolerances in knob-scaled lizards, after that of Berriozabal-Islas (2018). However, it is worth to mention that the referred author considered the organisms to reach their  $CT_{max}$  as the point when “they began to have an evasive behavior and tail whipping”. This criterion better matches the definition of voluntary thermal maximum ( $VT_{max}$ ; Taylor et al., 2021), and while its use is understandable as a less stressing, less invasive approach, it should not be confused with  $CT_{max}$ . That said, our results of  $CT_{min}$  and  $VT_{max}$  are similar to those documented by Berriozabal-Islas (2018) for *X. mendozai* ( $CT_{min} = 8.04^\circ\text{C}$ ;  $VT_{max} = 32.95^\circ\text{C}$ ), *X. tzacuaultipantecus* ( $CT_{min} = 8.81^\circ\text{C}$ ;  $VT_{max} = 32.26^\circ\text{C}$ ), and the population of *X. newmanorum* from Pisaflores ( $CT_{min} = 7.51^\circ\text{C}$ ;  $VT_{max} = 31.9^\circ\text{C}$ ). The population herein studied showed a narrow thermal tolerance range compared to other lizard species (Grigg & Buckley, 2013), and based on the similarity of our results and the above referred, it is likely a trait shared by all members of the genus *Xenosaurus*.

The observed degree of stenothermy of *X. newmanorum* is of utmost concern from a conservation perspective, as it implies an increased vulnerability to changes impacting the thermal environment of this lizard (Nowakowski et al., 2018). This, coupled with its microendemism, habitat specificity, low dispersal capabilities, and high rates of dehydration (Lara-Tufiño et al., 2013; Muñoz-Nolasco et al., 2019) suggest an elevated susceptibility for the species, in accordance with an assessment of extinction risk due to climate change previously conducted on Xenosauridae (Berriozabal-Islas et al., 2018). As thermoregulatory patterns might influence organismal response to climate change and even shift under different warming scenarios (Arenas-Moreno et al., 2021), it would be desirable to assess the potential of phenotypic plasticity in the thermal traits of these lizards. If confirmed, such plastic adjustments

could also help explain why some previous studies have found indications of active thermoregulation in *Xenosaurus* while others not (beyond methodological aspects). Whereas our study was limited in sample size and temporal spanning, the results are congruent with former research. Future studies should therefore look at describing more thoroughly the physiological ecology of knob-scaled lizards and determining the existence of active thermoregulation in other species of the genus.

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